REPORT DOCUMENTATION PAGE

AFRL-SR-AR-TR-05-

0281

1. REPORT DATE *(DD-MM-YYYY)*	2. REPORT TYPE	3. DATES COVERED *(From - To)*
07/07/2005	Final report	04/16/2004 - 07/07/2005

4. TITLE AND SUBTITLE
On-Chip Hardware for Cell Monitoring: Contact Imaging and Notch Filtering

5a. CONTRACT NUMBER
FA9550-04-1-0449

5b. GRANT NUMBER

5c. PROGRAM ELEMENT NUMBER

6. AUTHOR(S)
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5d. PROJECT NUMBER

5e. TASK NUMBER

5f. WORK UNIT NUMBER

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8. PERFORMING ORGANIZATION REPORT NUMBER

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Arlington, VA 22203

10. SPONSOR/MONITOR'S ACRONYM(S)

11. SPONSOR/MONITOR'S REPORT NUMBER(S)

12. DISTRIBUTION / AVAILABILITY STATEMENT

Approve for Public Release: Distribution Unlimited

13. SUPPLEMENTARY NOTES

14. ABSTRACT

The cell canary system will combine live bio-engineered cells with on-chip MEMS hardware. It will function by monitoring the response of many live cells to an external environment. Each cell, or cell colony, will be bio-engineered to fluoresce in response to a specific external pathogen. The fluorescence will occur through a FRET interaction with a quantum dot that labels a component of the pathway. If the cell or cell colony dies due to the presence of its target pathogen, then it will fluoresce as it dies; if the cell or cell colony dies due to other factors, then it will not fluoresce. The system will therefore act as a "cell canary" system for specific, low-false-positive detection of pathogens. Our goal here is to demonstrate proof-of-concept for the on-chip fluorescence detection portion of the project.

15. SUBJECT TERMS

16. SECURITY CLASSIFICATION OF:			17. LIMITATION OF ABSTRACT	18. NUMBER OF PAGES	19a. NAME OF RESPONSIBLE PERSON
a. REPORT	b. ABSTRACT	c. THIS PAGE		6	19b. TELEPHONE NUMBER *(include area code)*

7-13-05

Organization: University of Maryland

DSO Simbiosys

Title: On-Chip Hardware for Cell Monitoring: Contact Imaging and Notch Filtering

Start Date: 04/16/04 **End Date:** 06/07/05

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Project Goals

The cell canary system will combine live bio-engineered cells with on-chip MEMS hardware. It will function by monitoring the response of many live cells to an external environment. Each cell, or cell colony, will be bio-engineered to fluoresce in response to a specific external pathogen. The fluorescence will occur through a FRET interaction with a quantum dot that labels a component of the pathway. If the cell or cell colony dies due to the presence of its target pathogen, then it will fluoresce as it dies; if the cell or cell colony dies due to other factors, then it will not fluoresce. The system will therefore act as a "cell canary" system for specific, low-false-positive detection of pathogens. Our goal here is to demonstrate proof-of-concept for the on-chip fluorescence detection portion of the project.

Technical Approach

We will begin demonstrating proof of concept for the ability to fabricate optical filters on top of CMOS photodetectors that can block the excitation wavelengths but pass the emitted fluorescent light. We shall also continue development of "cell clinic" microstructures.

Major Accomplishments

- CMOS optical detectors were designed, tested for spectral sensitivity, and post-processed for optical selectivity to long wavelength light. Optical filters were designed, and chips were coated by commercial vendor with 49-layer interference filter comprising alternating layers of different index of refraction materials to block excitation light. Interference filters were also fabricated in-house by HDPECVD (37 layers), and measured characteristics are similar to the commercial coating. Absorption filters also designed and fabricated. Current generation of absorption filters uses PDMS as a polymer carrier. Spectrophotometer chosen and purchased for testing optical filters and materials. Characterization and comparison of fabricated absorption and interference filters is underway.
- Proof of concept demonstrated for integrated detection of fluorescence from quantum dots using straight-through optical path, down to 5nM concentration. Test setup excited quantum dot sample using a UV LED; excitation light plus emitted fluorescence passed through macroscopic low-pass optical filters and onto CMOS optical detectors.
- Fabrication of MEMS devices on CMOS chips continues. MEMS devices for 24-vial device designed. Robust MEMS fabrication process developed and devices demonstrated in high yield (>90%) and with excellent reproducibility of behavior. Multi-level SU8 process developed. Optimization of actuator for closing vial lids and development of lid sealing technology is underway: PPy/Au bilayer hinge bending angles characterized as a function of layer thicknesses for the full spectrum of achievable thicknesses; PPy/Au bilayer hinge force measurement protocol developed and mask to test forces designed and submitted; Hinge force measurements begun; PPy/Au bilayer hinge bending angles characterized as a function of temperature in NaDBS solution.
- Photopatternable polymers are a viable interim packaging solution; through-chip vias are under development as a long-term packaging solution. Intermediate methods such as molds and "chip caps" abandoned due to complexity and availability. Low swelling photopatternable polymers have been

identified and tested (*Loctite*® 3340, 3525, 4304). Back side DRIE of CMOS chips in order to implement "chip vias" is under investigation. A via design for back-side electrical contact has been developed, and first generation prototypes have been successfully fabricated on test wafers. Biocompatibility of the packaging materials has been established.

- Dielectrophoresis methods are being developed to provide robust, readily integrated technology for manipulating micro-scale particles in 3-D over heterogeneous surfaces and in biocompatible environments. Numerical simulations of DEP forces in FEMLAB agree well with experimental results obtained using particle imaging velocimetry. Novel techniques developed using negative and positive DEP simultaneously and sequentially.
- Potentiostat chips for in situ control of PPy/Au bilayer actuators have been designed, fabricated, and tested. The reference, working and counter electrodes have been integrated on the chip surface. All electrodes have been electrolessly plated with gold. Bench tests indicate successful operation of the on-chip potentiostat circuit using an off-chip Ag-AgCl reference, PPy-Gold working and graphite counter electrodes.
- Imaging chips have been designed, fabricated, tested on the bench, packaged using a photopatternable polymer, and tested with cells cultured on the chip surface. The contact imager is capable of resolving small clusters of stained cells.

DOD Impact

Enable reliable, low-false positives bio-chem detection system.

Technology Transfer/Products

B.Shapiro, E.Smela, P.Abshire, D.Wirtz "Cell Sensor Based Pathogen Detection". Provisional patent filed by OTC office Sept 2004. [Awarded Invention of the year in 2004 at the University of Maryland within the Physical Science category. One winner was chosen in each of the six categories by the Office of Technology Commercialization and by an external review process.]

Team Member Organizations

University of Maryland at College Park.

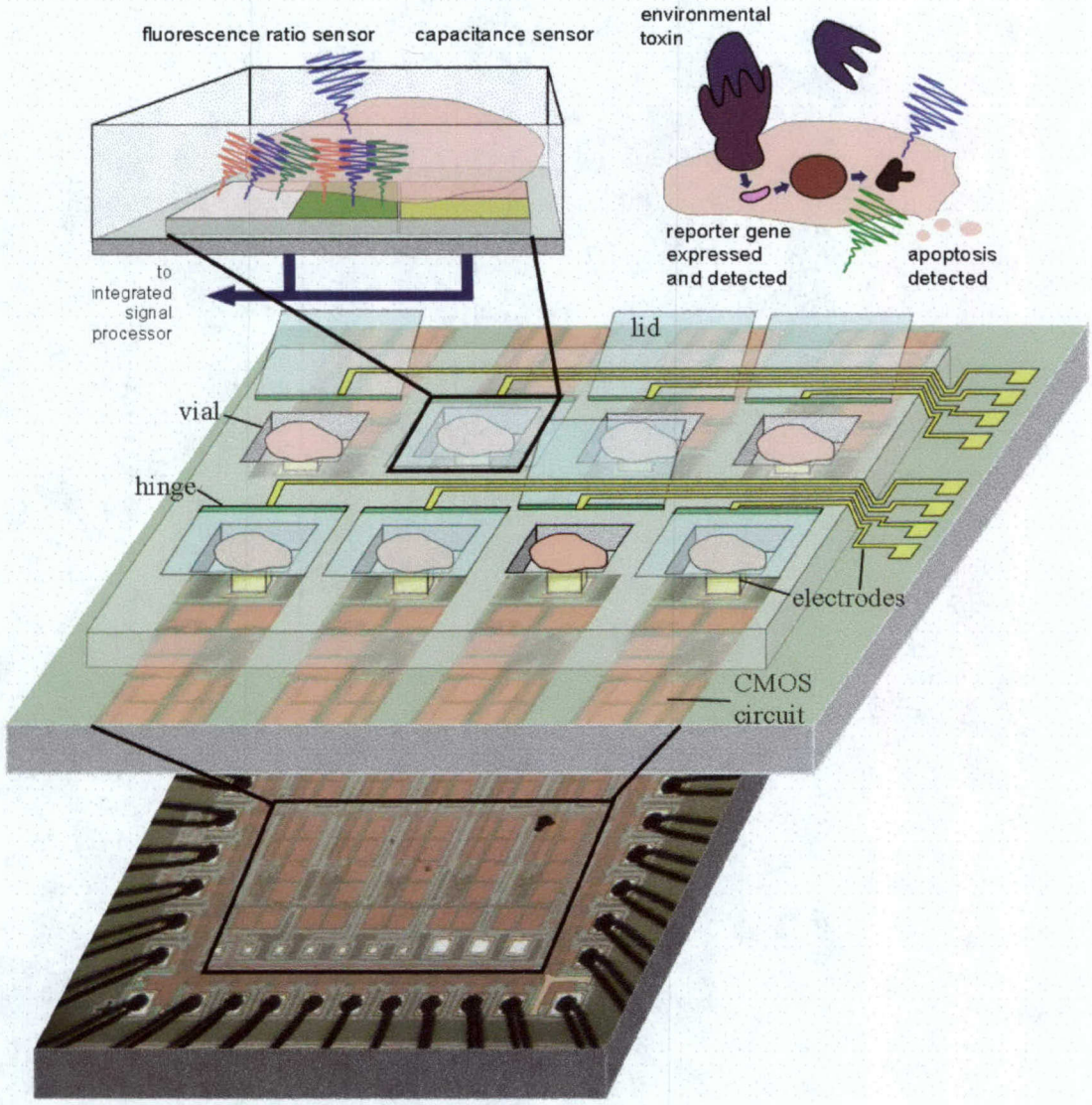


Figure 1: The concept for the "cell canary" chip. MEMS vials hold cells; cell life within each vial is monitored by CMOS circuitry that detects electrical and optical signals from the cells.

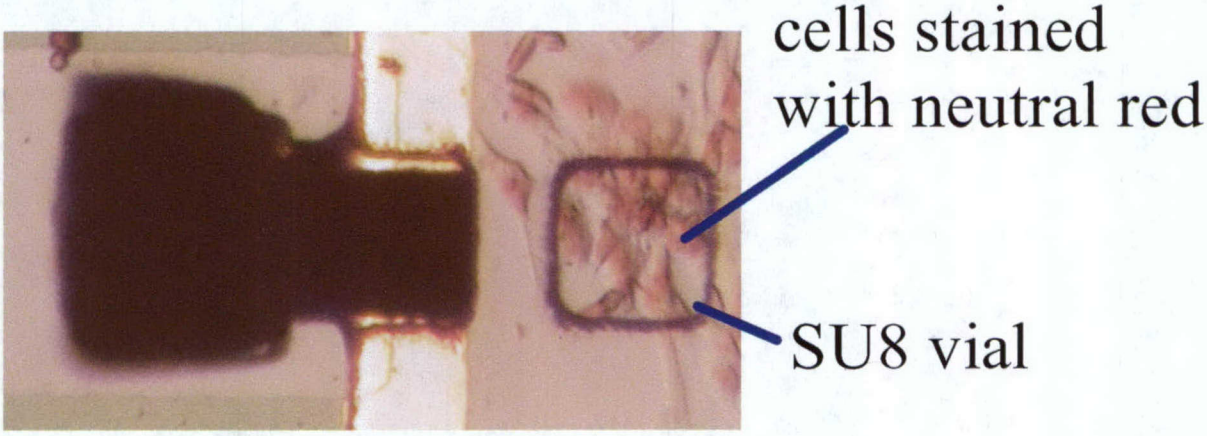
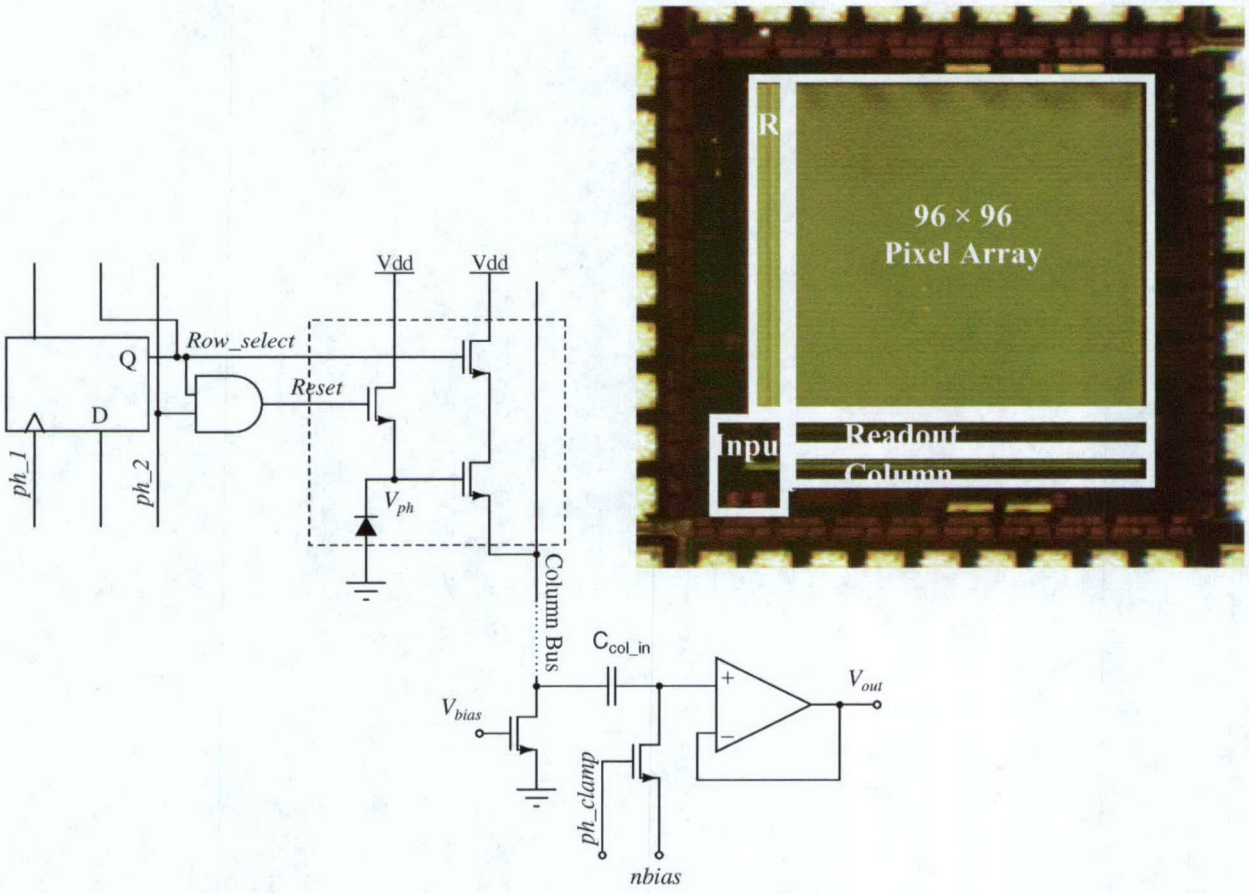


Figure 2: Bovine aortic smooth muscle cells (BAOSMC) stained with neutral red and cultured on the microstructures. Cells are clearly visible on the SU8 surface and inside the vial.



Process	AMI05
Maximum signal	1.2 V
Conversion gain	22 μV/e
Measured pixel noise	σ = 2.5mV over 2 ms integration time
Dynamic range	53.6 dB
Dark signal	0.46 V/sec
FPN (Fixed pattern noise)	3.18 mV

Figure 3: Contact imager comprises 96x96 pixel array with control and readout circuitry. Upper left, pixel circuit, controls, and correlated double sampling readout circuit. Upper right, photomicrograph of the imager. Bottom, measured characteristics of the imager.

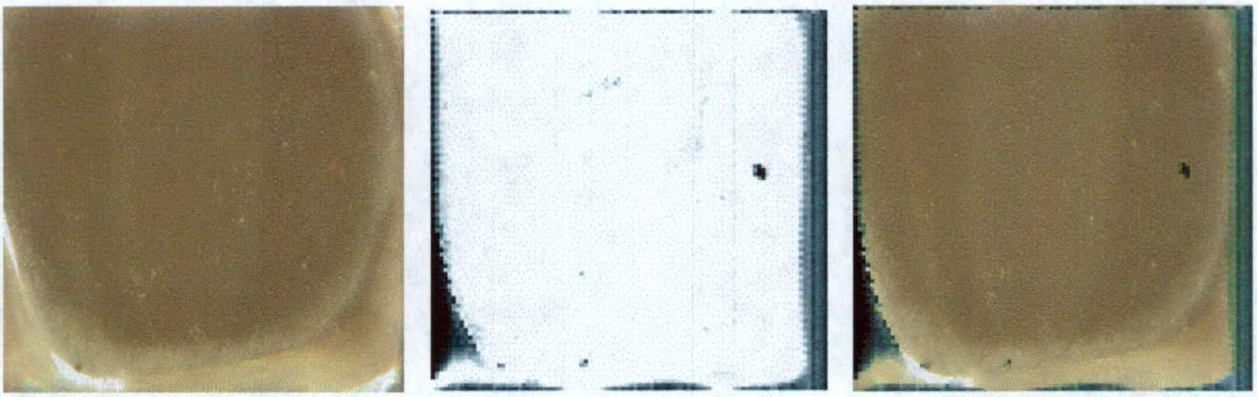


Figure 4: First generation contact imager tested in vitro with BAOSMC cells.

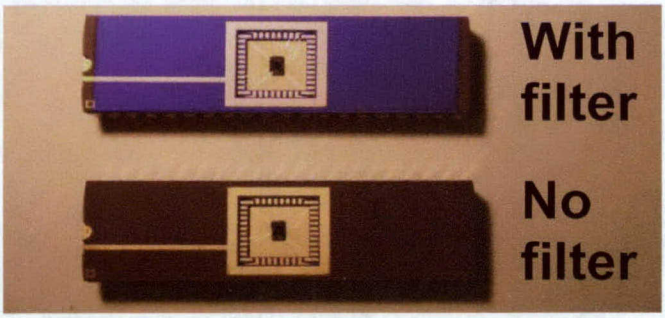


Figure 5: Optical detector chip post-processed and coated with interference filter. Alternating layers of different index of refraction materials block excitation light.

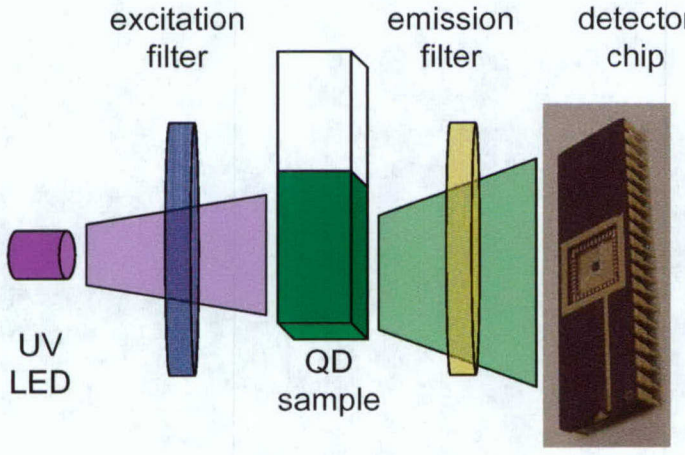


Figure 6: Experimental setup for detection of quantum dot fluorescence.

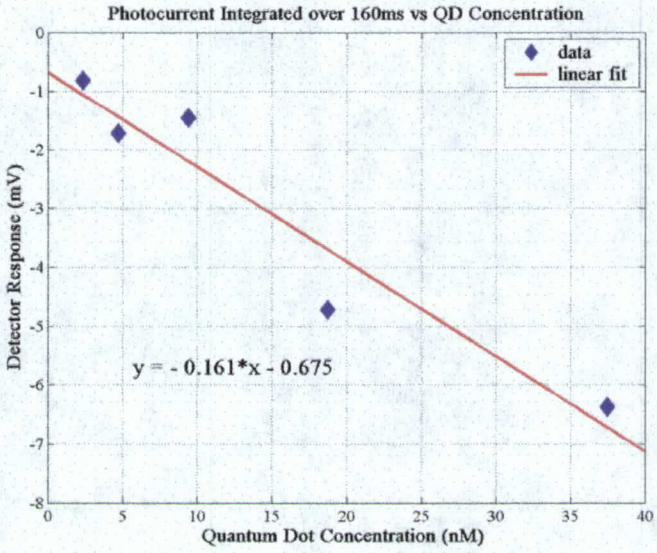


Figure 7: Proof of concept. Integrated photosensor measures quantum dot concentration down to 5nM concentration.